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A BACTERIAL DISEASE OF THE SUGAR BEET.¹

CLARA A. CUNNINGHAM.

(WITH PLATES XVI-XX)

IN the autumn of 1890 Professor H. A. Huston, chemist of the Indiana Experiment Station, noticed that the analyses of some sugar beets showed a much lower per cent. of sugar than others, and the difference seemed to be associated with a slight change from the usual appearance of the tissues of the root. This observation led to a microscopical examination of the affected beets by Dr. J. C. Arthur, who discovered the presence of bacteria in the tissues, to which, after further study, was attributed their abnormal condition. During the year 1891-2 the characteristics of the disease were studied by Dr. Arthur and Miss Katherine E. Golden, and the results published in the form of a bulletin in 1892.²

This preliminary series of investigations determined that the disease was associated with a specific germ, which could be readily isolated from the diseased tissue.

No similar disease of the beet had been reported from any other locality in America at the time of the publication of this work. Dr. Ernst Kramer, in 1891, reported a bacterial disease of the beet root attacking the fodder beets of Russia, and almost simultaneously Dr. Paul Sorauer, of Germany, reported a disease of the sugar beet of that country. In the *Export* of 1894,³ Dr. Sorauer gives his opinion that the disease of the fodder beet, named by Kramer "bacteriosis gummosis," and that of the sugar beet similarly named by himself "bacteriose gummosis,"

¹ Read before the Society for the Promotion of Agricultural Science at the Boston meeting, August 1898.

² Diseases of the sugar-beet root. Purdue University Agric. Exper. Station, Bulletin no. 39.

³ Export, 1894, no. 30.

are identical, and, perhaps, very closely related to the bacterial disease of sugar cane known as "sereh."

The diseased beets, as observed in Russia, are described as having dried leaves with withered heart leaves. The roots of badly diseased beets were so tough they could scarcely be broken, the broken surfaces soon turning black. These beets produced a pathogenic effect on cattle to which they were fed. Many of the diseased beets, when first sectioned, appeared perfectly sound, but after a few minutes the fibrovascular bundles turned dark and a syrup-like gum exuded from the cells. In other beets the tissue was sometimes completely broken down.

Dr. Sorauer says: "The similarity between the beet and the sugar-cane disease 'sereh' consists in the destruction of the cane sugar and the increase of the invert sugar as well as in the coloring of the vascular bundles and the entrance of bacteria." He also believes that the disease discovered in America by Arthur and Golden may be the same as that determined by Kramer and himself in Europe.

Mr. Walter Busse, in 1895, took up anew the study of the *bacteriosis gummosis* of the sugar beet, the material for study being sent him by Dr. Sorauer. In describing the diseased root he speaks of the gum-like fluid as follows: "Soon after the drops appear on the surface of the sectioned beet they are covered by a thin black membrane, which consists of small, black, round bodies of different sizes."

The aim of Mr. Busse was, first, to determine the form of bacterium common to all the diseased beets by the separation of the germ from the diseased tissue; and, second, to demonstrate that this germ was the specific cause of the disease by inoculating healthy beets with the germ. In the first series of experiments three germs were isolated. Two of these were discarded and the third form was kept for further observation. This form appeared as short rods $1.72-2\ \mu$ long and $0.8\ \mu-0.9\ \mu$ broad. They were actively motile, and grew well in cane or grape sugar solutions, producing an abundance of gas. This form was lost, but from other diseased beets a second gas-producing form was

isolated, resembling the first in form and arrangement, but smaller, being $1.5\text{ }\mu$ – $1.75\text{ }\mu$ long and 0.7 – $0.8\text{ }\mu$ broad. This form produced an acid reaction on different media, did not liquefy gelatin, and grew better at 12° – 14° than at 22° . Mr. Busse is inclined to believe that the second form is a variety of the first, which he designates as *Bacillus Beta*. He has demonstrated that this second form produces the disease known as *bacteriosis gummosis*, and believes that this germ is a saprophyte which becomes a parasite in the tissues of the beet.

Erwin F. Smith,⁴ in speaking of the bacterial diseases of the sugar beet as reported from Europe and America, is of the opinion that the diseased condition of the beets studied by Arthur and Golden is due to some other cause than a bacterial one. He states that it is highly improbable that the root could be attacked by an organism which invades its tissues and yet does not break them down. Mention is made of the fact of the presence of small bodies in the tissue of healthy beet roots which have the appearance of bacteria, but which are probably crystalloid bodies. A paper by Dr. Smith was presented at the meeting of the Society for Plant Morphology and Physiology in December 1897, calling attention to the "existence, in parts of the United States, of a disease of the sugar beet resembling if not identical with that described by Kramer and Sorauer in 1891–2, and more recently by Busse."

In the fall of 1896 I had the opportunity to continue the investigation of the bacterial disease of the sugar beet observed in Indiana in 1890–1. Much of the value of my experimental study of this disease is due to the suggestions of Dr. Arthur, to whom I am indebted for kindly help and criticism of my work. I also desire to express my gratitude to Professor Burrage, Professor Huston, Miss Golden, and Mr. H. L. Bryan, also of Purdue University, for important suggestions. My investigations, which have been continued from 1896 to the present time, have resulted in no positive evidence that the sugar-beet disease of Indiana is the same as that described by Sorauer and Busse

⁴SMITH E. F.: Am. Nat. 30:716–729. Sept. 1896.

of Europe. The points of similarity will be noted in the following description of the disease and of the germ by which it is produced.

GENERAL DESCRIPTION OF THE DISEASE.

About the middle of September 1896 several diseased beets were found in the field of cultivated beets on the grounds of the Purdue Experiment Station. The disease attacks the whole beet plant, causing a peculiar appearance of the leaves, so that with a little practice the diseased beets can be distinguished readily from the healthy ones as they grow in the field. The outer, older leaves soon die away, and the intermediate and heart leaves are left wrinkled, curled, rather flabby than turgescent, and of a yellowish-green color. This wrinkled appearance is caused by blister-like patches being formed between the veins of the leaf, and the whole has been described as resembling a Savoy cabbage leaf. See photographs of leaves, *plate XVI*, and also photographs of diseased beets, *plates XVII*, and *XVIII, A*.

The appearance of the exterior of a beet root when diseased is not materially different from that of the healthy beet. It is perhaps not quite as brittle. A decisive test for the disease is found in the appearance that the root shows when sectioned. The fibrovascular bundles appear as dark rings in the white flesh. They grow almost black after being exposed to the air for a few minutes. These rings are quite distinct from the cream colored fibrovascular bundles of healthy beets (*plate XIX*).

In 1896 in a field of beets covering an area of about one acre and containing approximately 130,000 beets, eleven badly diseased and several slightly affected ones were found. This was a smaller number than had been found on the same ground in previous years, and can perhaps be accounted for by the climatic conditions being so favorable to plant growth the preceding summer, there being an abundance of rain. The number of diseased beets increased, however, toward harvest time.

Frost seems to be much more injurious to the diseased than

to the healthy beets. The heart leaves of the diseased beets were more easily injured by the frost. It is characteristic of the disease that the leaves of badly diseased roots die away until no green leaves remain, leaving an apparently dead root in the soil, though its tissues will be found to be firm and not in the least broken down. The early frosts hasten the destruction of the leaves. Both diseased and healthy roots show an acid reaction, the diseased seeming slightly more acid than the healthy.

ETIOLOGICAL HISTOLOGY.

Comparative study was made of sections taken from both diseased beet roots and healthy beet roots, also sections from leaf and leaf-stalk of both diseased and healthy beets. In all these sections small round bodies were seen in the cell substance. These bodies were found to be protein by their reaction to iodine. They measured from $2-4\mu$ in diameter and turned yellow when treated with iodine. In the tissues of diseased beets other bodies were found which were smaller and of a different refractive power and arrangement. These bodies stained in gentian violet like bacteria, and looked almost like micrococci when imbedded in the cell substance, but when free in the water were easily distinguished as small motile bacilli.

SEPARATION OF THE SPECIFIC GERM.

The first steps in the separation of the germ were as follows. A diseased beet was selected, a thin knife sterilized in the flame and used to remove all parts of the beet exposed to the air. A small piece of beet was then removed from the heart of the root and transferred by means of a sterilized platinum wire to tubes of melted gelatin or agar. The first series of cultures was made by inoculating gelatin tubes with pieces of diseased tissue, at the same time inoculating a number of tubes in the same manner with pieces of healthy beet as a control. The following tables give the results of a series of such inoculations. The first shows the results of cultures made from diseased tissue; the second the results of cultures made from healthy tissue.

TABLE I.

CULTURES ON ARTIFICIAL MEDIA FROM DISEASED AND
HEALTHY BEETS.

| Dates and media | Number of cultures | Results | |
|-----------------------------|--------------------|----------------------------------|-------------------------|
| FROM DISEASED BEETS. | | | |
| Sept. 20 Gelatin - - | 10 | Oct. 5 Characteristic growth | Oct. 10 Contaminated |
| Oct. 2 Gelatin - - | 3 | Oct. 4 Characteristic growth | Dec. 10 Still pure |
| Oct. 24 Glycerin gelatin | 4 | Oct. 28 Characteristic growth | |
| Oct. 24 Glucose gelatin | 4 | Oct. 28 Characteristic growth | |
| FROM HEALTHY BEETS. | | | |
| Sept. 20 Gelatin - - | 4 | Oct. 5 No growth | Oct. 10 No growth |
| Oct. 2 Gelatin - - | 3 | Oct. 4 ⁵ No growth | Dec. 10 No growth |
| Oct. 24 Glycerin gelatin | 4 | Oct. 28 No growth | |
| Oct. 24 Glucose gelatin | 0 | | |

All the above cultures were made in standard gelatin not titrated or in standard agar to which had been added 5 per cent. glycerin or 5 per cent. glucose. The growth in successful inoculations was the same in all cases. Some creamy-white globules grew out on either side of the diseased tissue, and in the course of a few days were surrounded by a lens-shaped capsule or break in the gelatin caused by the gas that was given off in the growth. When the bubbles reached the surface the growth was distributed in white rings around the tube (*plate XX, B"*).

October 15 several cultures were made in standard gelatin. In some of these a growth resulted but no successful transfers were made. One of these cultures was used to inoculate a healthy beet in the field.

⁵ Photograph of tubes at this stage shown in *plate XX*.

November 3 stab cultures were made from a tube of glycerin agar inoculated with diseased tissue. These were all contaminated with the exception of one which formed a perfectly colorless layer, gelatinous in consistence, on agar and sterilized beet. This form will be spoken of later.

No growth resulted from a series of cultures made in 10 per cent. cane sugar gelatin.

In December a diseased beet, which was frozen the previous night, was brought in from the field. Pieces of this tissue were transferred with the usual precautions to tubes of melted gelatin to which had been added 5 per cent. of cane sugar. The growth in these cultures was rapid and gas was given off in large quantities. Stab cultures were made from these and appeared exactly uniform, and just the same in appearance as stab agar cultures made directly from unfrozen diseased beet, with one exception in each case. From one of these exceptional tubes the perfectly colorless gelatinous form spoken of above was found. This form was also obtained in one of the stabs taken directly from the diseased beet.

In the above inoculations the growth was much more rapid than in previous cultures, probably because the tissues were broken down by freezing so that the germ could escape more easily into the surrounding medium.

Another series of cultures was made at the same time from a frozen beet in which the disease was produced by inoculation. The growth was similar to that of the preceding series. Transfers taken from these were uniform and similar to those described above.

In these inoculations a plug was removed from the healthy beet root with a sterilized knife, the inoculating material inserted, the plug replaced and covered with cotton. The table on the next page shows the results of inoculations of three beets in the field.

The beet inoculated with diseased tissue was slower in showing the disease than the one inoculated with the germ growing on gelatin, probably because of the time required for the germ to make its way through the cell walls of the tissue.

TABLE II.

RESULTS OF INOCULATING BEETS IN THE FIELD.

| Beet | Date of inoculation | Source of germ | October 22 | November 19 | November 28 |
|------|---------------------|-----------------|-----------------------------------|-----------------------|------------------------|
| 1 | Oct. 17 | Isolated germ | Yellowish color of leaves noticed | Disease quite evident | Removed from the field |
| 2 | Oct. 17 | Diseased tissue | Slight indication of the disease | Disease quite evident | Removed from the field |
| 3 | Oct. 17 | Healthy tissue | No change | No change | No change |

The heart leaves of the inoculated beets showed the effect of the disease in the slight blister-like areas on their surfaces. The beet inoculated with the gelatin containing the germ, and the one inoculated with healthy tissue, were brought into the laboratory November 28, as the progress of the disease could not be followed out of doors because of injury by frost. The beets were placed in culture jars of water in the greenhouse, where the healthy beet after some time decayed, and the diseased beet developed new leaves which were more or less crinkled and faded, but gradually assumed a smoother and darker green appearance, but the plant was still stunted in growth.

INOCULATIONS IN THE GREENHOUSE.

The method of inoculation of beets in the greenhouse was similar to that of inoculation of beets in the field. As a result of these trials several beets seemed to show the effects of the disease to a slight degree.

DESCRIPTION OF THE GERM.

The germ as isolated from the diseased tissue is a small bacillus measuring from $0.9-1.0-1.3 \mu$ in length, and $0.5-0.8 \mu$ broad. When taken from the culture media the germs are arranged singly or in pairs, and possess individual motion. The germ

seems to revolve more or less irregularly on its axis. The germ stained well with all the common bacterial stains. No process of staining showed the presence of spores or flagella (*plate XX, B*).

EFFECT OF LIGHT ON GROWTH.

The germ grew better in the dark than in the light. Germs taken from old, dried out cultures were smaller than when grown on a moist substratum. Desiccation also injures the capability of the germ for motion. Germs taken from an old culture and examined in a drop of water were less motile than those taken from a fresh bouillon culture.

The germ grows better at a temperature of 12° - 14° than at 21° . Stab cultures in agar grew slowly at body temperature. The germ grown in bouillon and exposed to a temperature of 100° for five minutes was killed.

GROWTH OF THE GERM ON DIFFERENT CULTURE MEDIA.

Stab cultures of gelatin showed a thin grayish-white layer on the surface, and extending down the line of inoculation. As the cultures grew older the color darkened to a deep cream. The gelatin was liquefied in the course of several weeks. When melted gelatin was inoculated and then allowed to solidify there was a growth throughout the gelatin in streaks and films. Colonies on gelatin plates were not distinctly outlined and were sometimes accompanied by a disagreeable odor. The germ seemed to grow better on agar than gelatin. Agar to which had been added 3 per cent. of cane sugar or glucose seemed to specially favor its growth. The growth on slant agar was drab-white in color, smooth margins, and a slow and not luxuriant growth. The growth was not viscous.

In agar plates the colonies have their origin in the deeper layers of the agar where they are generally elliptical. When they reach the surface they spread out in their round grayish-white colonies with compact creamy-white centers. In bouillon growth is observed after two or three days. No turbidity of the fluid was observed, but a sediment was deposited in the bottom

of the tube. Masses of zooglœa were sometimes found in old bouillon cultures. The germ grew well on sterilized sugar beet, and deposited a sediment in sterilized beet juice. The germ grew on sterilized apple, potato, and turnip. A raw potato was broken open and inoculated with the germ. There was a slight growth developed. A raw sugar beet was inoculated. The germ grew, causing a black coloration of the fibrovascular bundles, and in a microscopical examination was seen to have entered the tissue.

NITRATE SOLUTION.

This solution was prepared using 1000cc distilled water, 1 gram peptone, and 1 gram potassium nitrate. Tubes of this solution in which the germ had grown for three days when tested showed that the nitrate had been fairly well reduced.

ACID AND ALKALINE MEDIA.

It has been stated that the beet root is acid to the extent of little over 1 per cent. Because of this fact experiments were made with acid and alkaline media in order to determine which of the two would be more favorable to the growth of the germ. In bouillon, to which had been added 1 per cent. malic acid, the solution was made neutral; 5 per cent. acid solutions were not rendered neutral. The solution was not made turbid by the growth of the germ. In 1 per cent. alkaline solution of bouillon the germ was more motile than in 1 per cent. acid solution. In 3 per cent. alkaline solutions the germ was more motile and larger than when grown in acid media, measuring from 1.1-1.9 μ long, and 0.9-1 μ broad. In 5 per cent. alkaline media the sediment deposited was quite viscous. Zooglœa masses were found more or less abundantly in all the alkaline cultures. These solutions were not made acid in reaction by the growth of the germ.

STARCH SOLUTIONS.

In a solution composed of one part each of starch filtrate and bouillon, the germ grew but the starch was not reduced. These cultures were tested for starch and gave a decided

reaction ; they were also tested with Fehling's solution for glucose, but gave no reaction. The germ grew well in wort gelatin.⁶ When this gelatin was melted, and after being inoculated was allowed to solidify, the germ grew under the surface, producing bubbles of gas all through the gelatin.

Tests made of bouillon containing cane sugar in which the germ had grown gave a reaction for glucose. In order to determine if the enzyme existed outside the cell, a solution in which the germ had grown was filtered through a porous cup. This filtrate added to a 5 per cent. cane sugar solution, and tested with Fehling's solution for glucose gave no reaction. Further experiments are necessary before deciding definitely in regard to the enzyme properties of the germ.

CELLULOSE SOLUTIONS.

As the germ penetrates the cell wall of the plant in some way, experiments were made to determine its effect on cellulose. For this a special nutrient solution was prepared as follows :⁷

| | | | | | | | | |
|--------------------|---|---|---|---|---|---|------|-------|
| Distilled water, | - | - | - | - | - | - | 250 | cc. |
| Pepsin, | - | - | - | - | - | - | 2.5 | gram. |
| Magnesium sulfate, | - | - | - | - | - | - | .45 | " |
| Calcium phosphate, | - | - | - | - | - | - | .45 | " |
| Ammonium sulfate, | - | - | - | - | - | - | 2.05 | " |
| Sodium chlorid, | - | - | - | - | - | - | 1.25 | " |
| Beef extract, | - | - | - | - | - | - | 1.25 | " |
| Swedish filter. | | | | | | | | |

In this solution the growth was very slow, and a very small amount of gas was produced, and the cellulose slightly broken down.

FERMENTATION.⁸

Because of the abundance of gas produced by the germ in its growth, special fermentation solutions were prepared. The

⁶ Wort gelatin was made by adding 10 per cent. of gelatin to wort.

⁷ Sur la fermentation de la cellulose. Centralblatt für Parasitenkunde 2:358. 1896.

⁸ PAMMEL, L. H. and EMMA : A report concerning gases produced by bacteria in fermentation. Centralb. f. Parasitenkunde 2:707. Dec. 1896.

gas produced by the growth of the germ in these solutions was analyzed by Mr. H. S. Bryan, of Purdue University, the results of which are given in Table III. The tests were made for CO_2 , O, N, NH_4 , and CO, the difference being considered as hydrogen. In the first analysis a much larger amount of nitrogen was obtained than in any of the other cultures. The cultures for this analysis were several weeks old, and were perhaps not trustworthy.

There were some irregularities in the amount of gas produced, which cannot be accounted for. At one time 2 per cent. cane sugar bouillon containing no pepton when inoculated gave a large amount of gas, 20^{cc} being collected in each fermentation tube; 2 per cent. cane sugar bouillon containing no pepton, inoculated at another time under exactly similar conditions, gave only 2^{cc} of gas in each fermentation tube.

The germ grown in bouillon to which had been added 2 per cent. glucose at one time gave a very large amount of gas; at another time there was not enough gas produced to be analyzed. The gas produced by the germ as determined by analyses is composed of a very small amount of oxygen, less than 4 per cent., carbon dioxide approximately 44 per cent., nitrogen 17 per cent., and hydrogen approximately 30 per cent. Fermentation was produced in sterilized beet juice, Pasteur's solution, and maple sap. No fermentation was produced by the growth of the germ in bouillon to which no sugar had been added.

SUMMARY.

It has been determined that a microscopical examination of the tissues of diseased beets reveals the presence of bacteria in the cells of the plant. The tissues of the plant are not broken down, and the bacteria in all parts of the plant appear to be the same. Transfers of diseased tissue to the healthy beet root resulted in changed appearances of the plant which indicated almost certainly that the disease was transmitted.

The manner in which the germ finds entrance to the plant has not been determined. The conditions most favorable to the

TABLE III.
ANALYSIS OF THE GAS PRODUCED BY THE SUGAR BEET GERM.

| Date of Inoculation | No. of tubes | Solution | Amount of gas | Results of analysis | | | |
|------------------------|-----------------|---|-------------------------------|---------------------|---------|-------------------------|------------------------|
| | | | | CO ₂ | O | H | NH ₄ and CO |
| Dec. 18 | 3 | 5 % cane bouillon | Dec. 21. 4 ^{cc} | 25% | a trace | 50-60% | none |
| Jan. 18 | several | 2 % cane bouillon | Jan. 22. 20 | .5% | 20.6% | 11.9 | " |
| Jan. 25 | 3 | 2 % cane bouillon | Jan. 29. 15 | .69 | 27.5 | 10.1 | " |
| Jan. 25 | 3 | 2 % glucose bouillon | Jan. 29. 20 | 38.7 | 36.5 | 21.2 | " |
| Jan. 25 | 2 | 2 % lactose bouillon | Not enough gas to be analyzed | 3.6 | | | |
| Jan. 25 | 3 | 2 % cane bouillon, no pepton | Feb. 2. 20 | 37.63 | 7.22 | 21.64 | " |
| Feb. 3 | 7 | 2 % glucose bouillon | Feb. 11. 2 | 18 | 1.7 | explosion probably H | " |
| Feb. 3 | 3 | 2 % cane bouillon, no pepton | Feb. 11. 2 | No analysis made | 33.51 | | |
| March 29 | several | 5 % cane bouillon, with 3 % calcium chlorid | April 26. 10 | 44.23 | 1.92 | 33.65 | 14.02 |
| March 29 | several | 7 % acid wort solution | April 26. 15 | 53.27 | .9 | 31.77 | 14.09 |
| April 1 | several | Cellulose solution | Small | No analysis made | | | |

attack are those resulting from drought with succeeding low temperature.

The fact that the germ breaks down cellulose slowly explains the manner of its progress from one cell to another.

Experiments have shown that the germ in a medium of low per cent. acid grows nearly or quite as well as in one of alkaline nature, so that the acid element of the beet root does not offer material resistance to the germ.

The germ converts cane sugar to glucose in the process of producing gas. The amount of gas produced is not constant, but the reasons for this irregularity have not been determined.

The germ grows well with any form of sugar and especially well in media containing cane sugar. This fact makes it seem probable that the germ is especially at home on those media which contain sugar in some form, although it will keep alive on media without sugar, and after cultivation for a time on such media will adapt itself to the conditions presented.

ANOTHER ORGANISM SEPARATED FROM THE SUGAR BEET.

The colorless gelatinous form separated from the beet root in connection with the disease germ was at first thought to be an undescribed germ or rather the product of a germ, for only a few bacterial bodies could be detected under the microscope even when comparatively large masses of the substance were placed in the field. The organism appeared as small bacilli or micrococci.

The mass resembles the form of *Leuconostoc* so common in the vicinity of sugar refineries. Under the microscope, however, no streptococci were found, which characterizes *Leuconostoc* under the microscope. The gelatinous substance is soluble in water and alcohol; in the latter it turns to a milk-white substance before it dissolves. The substance increased rapidly in bulk when grown on sterilized beet. The mass did not dry out for months after the substratum had become dry and hard.

The substance grew well on 10 per cent. cane sugar agar. The growth was slow at first, but after a week or two masses

measuring a quarter of an inch in thickness and three fourths of an inch in circumference, collected on the surface of the medium in stab and slant cultures. In case of stab cultures the agar was broken vertically along the line of inoculation. The colorless growth followed this break in the agar, and as the substratum became hard the mass collected as a colorless semi-fluid in the bottom of the test tube.

On slant agar there was a thin colorless layer, imparting a fluorescent hue to the medium. In agar plate cultures the organism formed small round colonies about the size of a pin head, resembling a small drop of water. These colonies were sometimes found with the disease germ, in plate cultures taken directly from the beet. It also grew on sterilized potato, and to some extent on gelatin. Immediately after separation from the beet root the organism produced fermentation, but the power was lost after a time. Staining revealed only a structureless mass containing a few bacteria-like bodies.

Desiccation has little effect on the substance. Sections of beet on which the organism was growing have been kept in the laboratory until they are quite dried out, and the gelatinous mass is still apparent.

If this is indeed a form of *Leuconostoc*, it is interesting to find it in diseased beet roots.

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EXPLANATION OF PLATES XVI-XX.

PLATE XVI.

Leaves of healthy and diseased beets, as they appeared when brought in from the field. The three diseased leaves can readily be distinguished from the two healthy ones because of their blistered and crinkled surface.

PLATE XVII.

A diseased beet brought in from the field. The root is quite firm, none of the tissue being broken down. The leaves hanging down are quite dead and dry. The erect heart leaves are alive and show the characteristic crinkled surfaces.

PLATE XVIII.

A, a diseased beet with the dead leaves removed. *B*, a healthy beet of same size and stage of growth, also with the dead leaves removed.

PLATE XIX.

A, cross sections through the crown of healthy and diseased beet roots. *a*, healthy root. *b*, diseased root, characterized by the black rings of vascular tissue.

B, cross sections through the central portion of the same diseased and healthy beet roots. *a*, healthy root. *b*, diseased root.

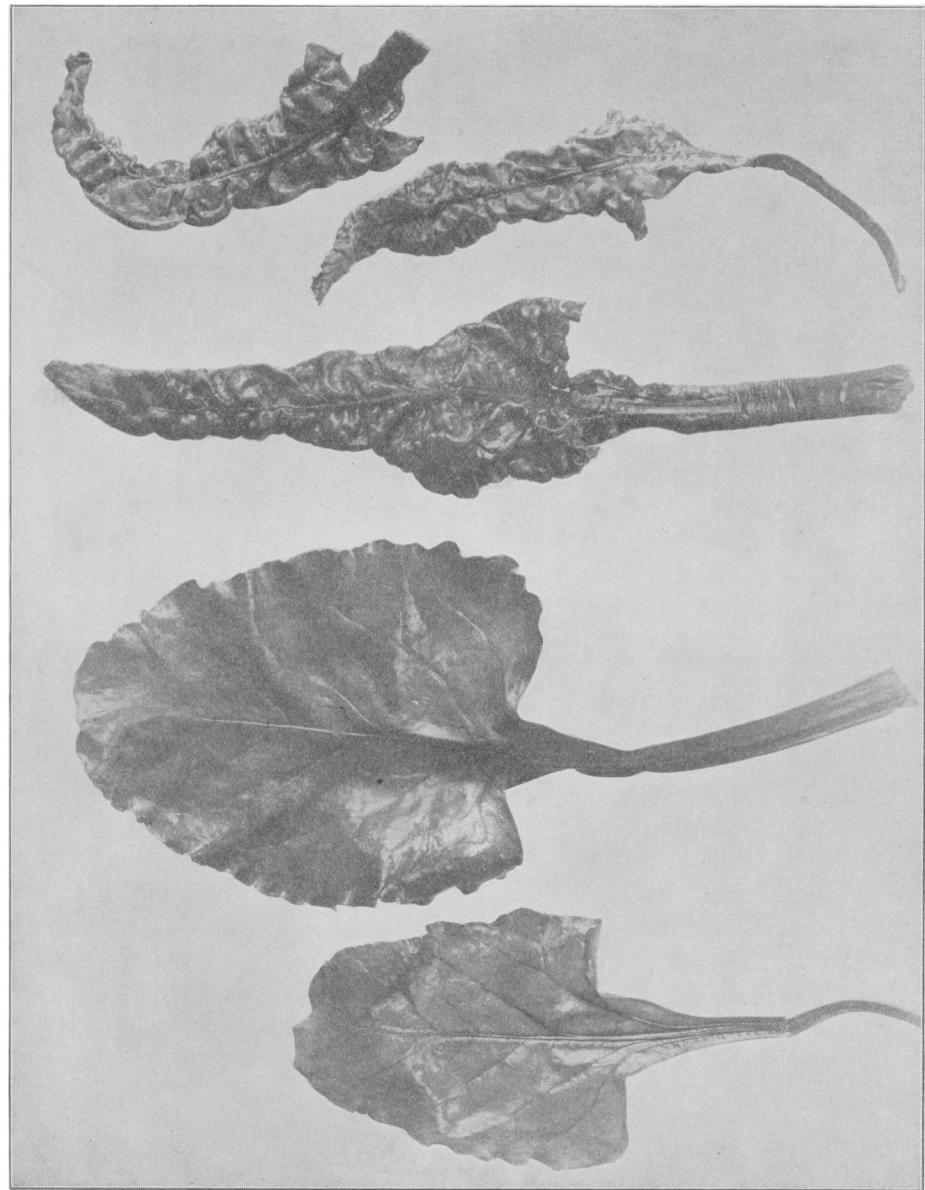
C, cross sections near the tip of the same roots. *a*, healthy root. *b*, diseased root.

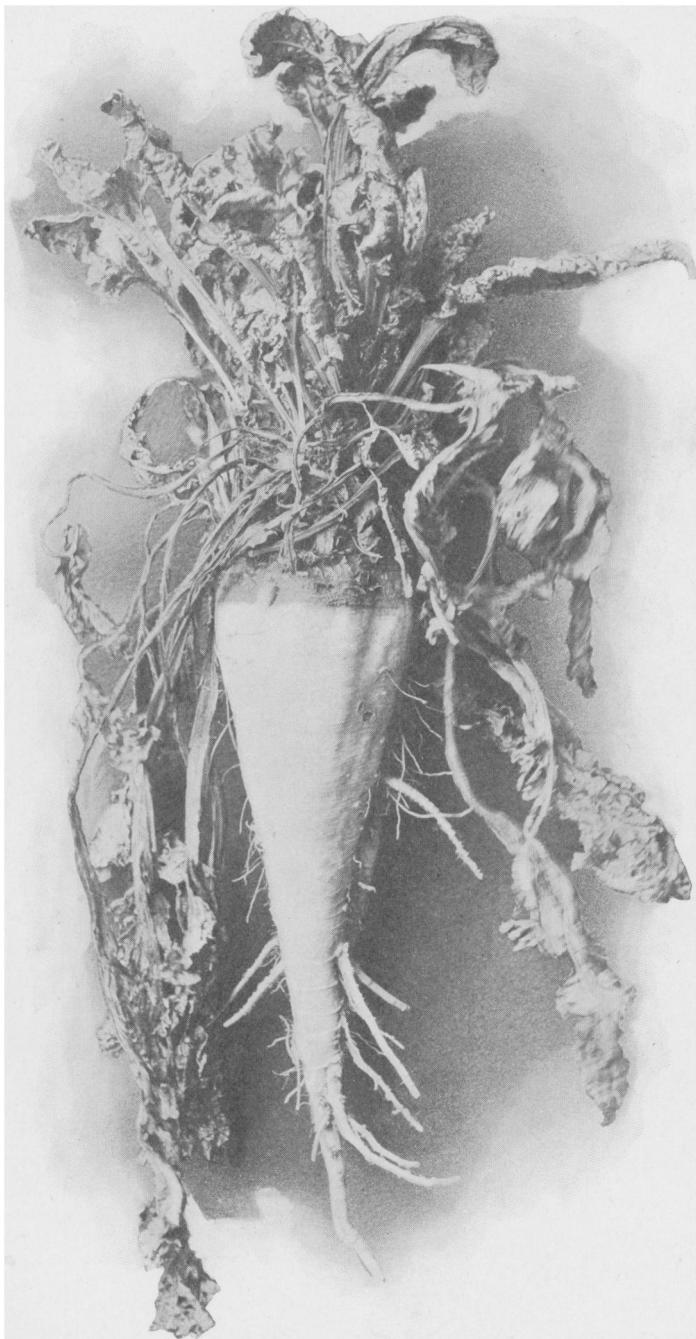
PLATE XX.

A, longitudinal sections of the same healthy and diseased beet roots figured in the preceding plate. *a*, healthy beet. *b*, diseased root.

A', tube of gelatin inoculated with a piece of healthy beet. *B'*, similar tube inoculated with a piece of diseased tissue. The photographs were taken after being inoculated two weeks. In *A* no growth appeared. In *B* the small globules of growth can be seen, breaking the agar and pushing it upward by the production of gas.

B, the disease germ stained with carbol fuchsin; imperfectly photographed.

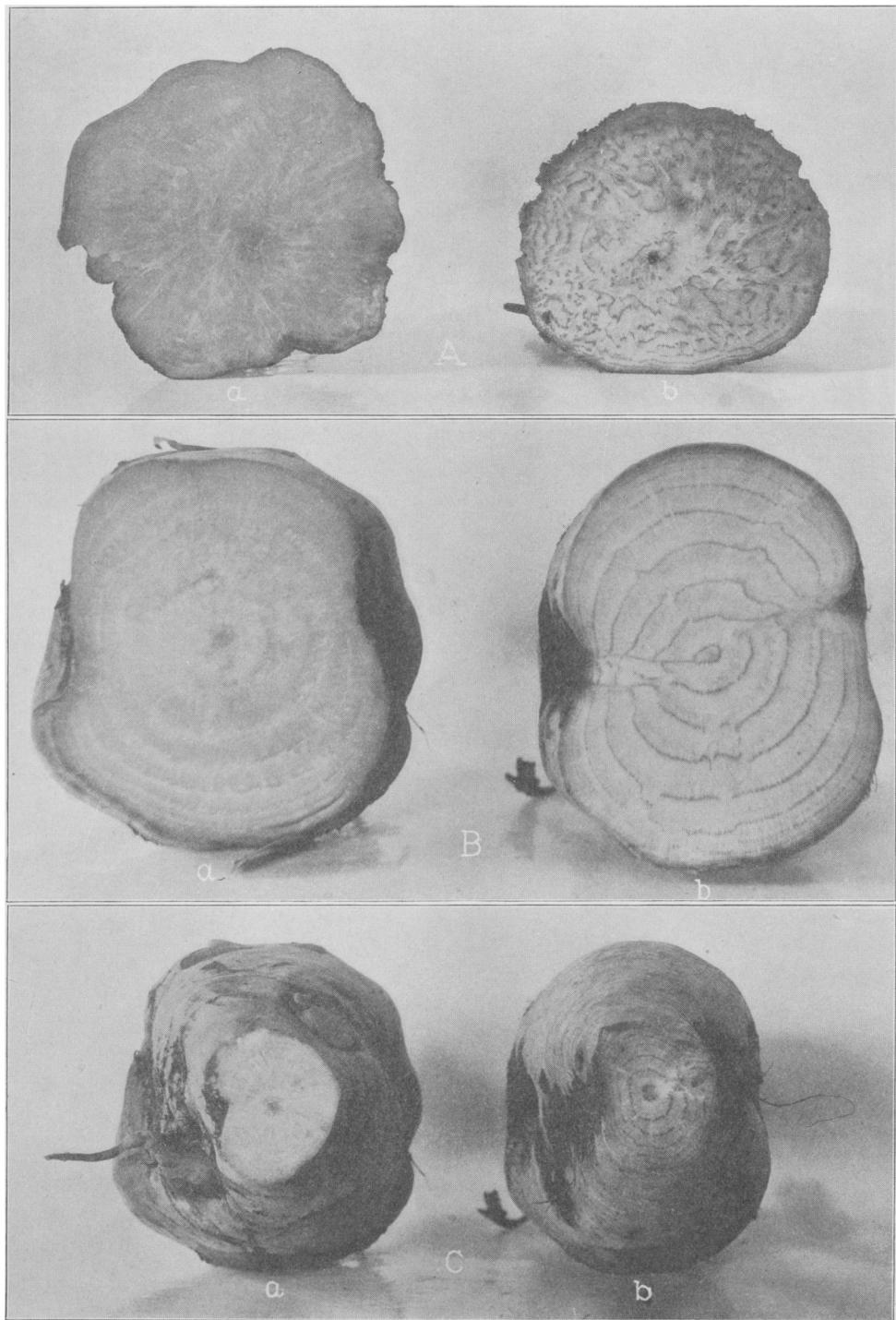




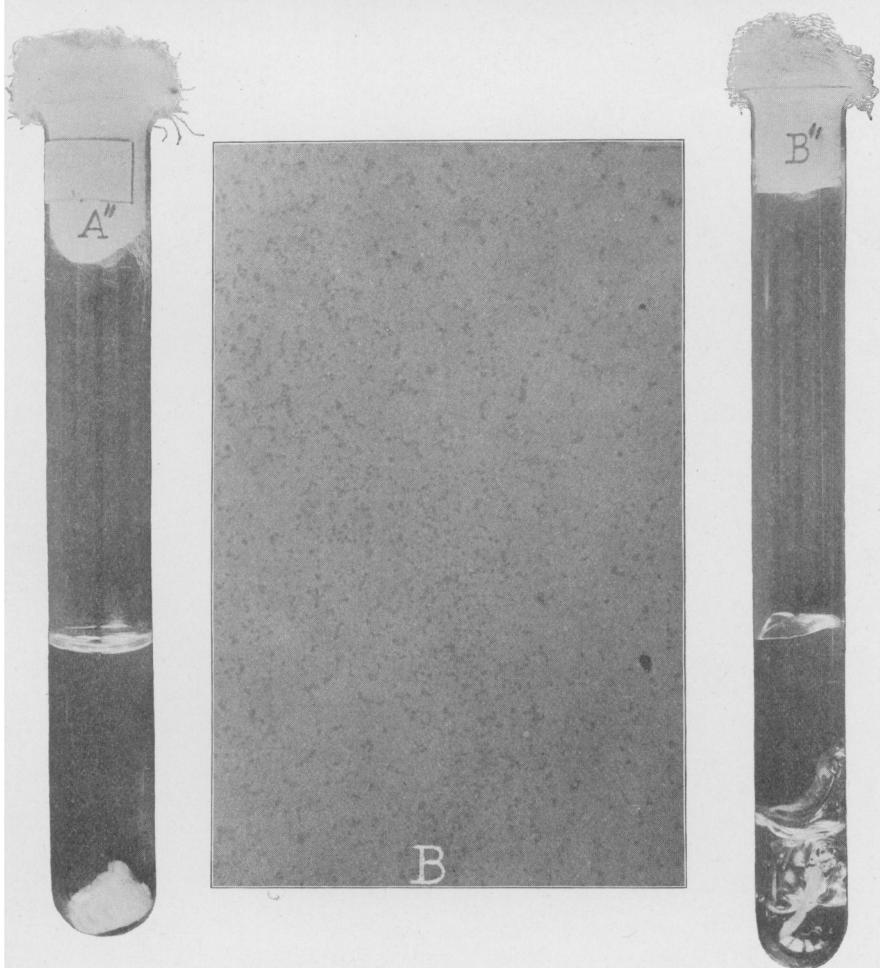
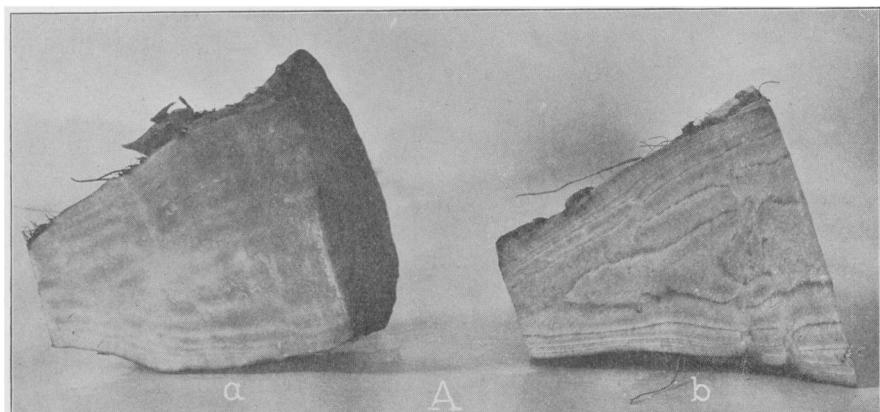
CUNNINGHAM on SUGAR BEET



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